

Research Note

Prevalence and Isolation of *Toxoplasma gondii* from Wild Turkeys in Alabama

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ABSTRACT: Hearts from 16 adult, male wild turkeys (*Meleagris gallopavo*) from eastern central Alabama were examined for encysted *Toxoplasma gondii* by hydrochloric acid–pepsin digestion and mouse inoculation. *Toxoplasma gondii* was isolated from 8 (50%) of the 16 wild turkeys examined. Six of the 8 isolates caused fatal infections in mice following primary isolation; a seventh isolate did not cause death, although the mice had clinical signs of infection prior to necropsy. Sera and/or tissue fluids from these 16 wild turkeys and an additional wild turkey were examined for *T. gondii* antibodies in the modified direct agglutination test. Twelve (71%) of the 17 samples were positive at dilutions of 1:50 or higher.

KEY WORDS: *Toxoplasma gondii*, tissue cysts, bradyzoites, tachyzoites, wild turkeys, *Meleagris gallopavo*.

Little is known about the prevalence or importance of encysted *Toxoplasma gondii* Nicolle and Manceaux, 1909, in wild game birds. Howerth and Rodenroth (1985) described a case of suspected fatal systemic toxoplasmosis in a female wild turkey (*Meleagris gallopavo*) from Georgia. The carcass had been frozen prior to examination at necropsy. Gross lesions consisted of splenomegaly, focal to confluent gray areas of consolidation in the lungs, and 1–3-mm-diameter erosions in the cecum and colon. Microscopic examination of tissue sections stained with hematoxylin and eosin indicated that protozoans were present in the brain, spleen, lungs, liver, adrenal glands, kidneys, esophagus, proventriculus, and colon. Ultrastructural examination of parasites in the lungs was suggestive of *T. gondii*. A direct fluorescent antibody test performed on a section of the spleen yielded results suggestive of the presence of *T. gondii*. Howerth and Rodenroth (1985) attempted to demonstrate *T. gondii* in mice inoculated with splenic tissues from the wild turkey but failed to isolate the parasite. Interestingly, Burridge et al. (1979) examined serum samples from 20 wild turkeys in Florida using the indirect hemagglutination test but failed to detect antibodies to *T. gondii* in any of the birds.

The present study was conducted to determine the prevalence of *T. gondii* in free-ranging wild turkeys and to attempt to isolate the parasite from their tissues. Experimental studies in domestic turkeys have demonstrated that the heart is most often infected with *T. gondii* tissue cysts and that the modified direct agglutination test (MDAT) using formalin-fixed tachyzoites will detect *T. gondii* antibodies in their sera (Dubey et al., 1993).

The hearts from 16 hunter-killed and sera/tissue fluids from 17 hunter-killed male wild turkeys were examined. The turkeys were from eastern central Alabama originating in Lee, Macon, and Russell counties. Hunters eviscerated the wild turkeys, placed the heart and viscera in a plastic food storage bag, and refrigerated the samples until they were delivered to the College of Veterinary Medicine, Auburn University. Blood and contaminating tissue fluid were collected from the plastic bags and centrifuged in a microfuge, and the supernatant was collected and examined in the MDAT for antibodies against *T. gondii* at dilutions of 1:25, 1:50, and 1:500 as described by Dubey and Desmonts (1987). Nineteen to 25 g of heart tissue was collected from each bird and digested individually in hydrochloric acid–pepsin solution (0.52 g pepsin, 0.5 g NaCl, 1.4 ml concentrated HCl, and 98.6 ml distilled water) and used for subcutaneous inoculations of groups of 4–5 female, 20–24-g Hsd:ICR mice. Four weeks later, serum was collected from all surviving mice and examined for IgG antibodies to *T. gondii* at dilutions of 1:50 and 1:100 in an indirect immunofluorescent antibody assay against RH isolate *T. gondii* tachyzoites as described by Lindsay et al. (1990). After serum was collected, the mice were killed, and squashes from their cerebrums were examined unstained for *T. gondii* tissue cysts as described by Lindsay et al. (1991). Smears were made from the lungs and/or cerebrums of mice that died during the study and examined as unstained preparations for stages of *T. gondii*.

Table 1. Serological prevalence and results of isolation of *Toxoplasma gondii* from wild turkeys from Alabama.

Turkey	Heart*	NI/NP/NPD†	% mortality‡	MDAT§
1	NA	NA/NA/NA	NA	1:50
2	25	5/5/1	20	1:50
3	25	4/4/4	100	1:50
4	25	4/4/1	25	1:50
5	25	4/4/1	25	1:50
6	21	4/0/0	0	<25
7	19	4/0/0	0	1:50
8	25	5/0/0	0	1:50
9	25	4/0/0	0	1:50
10	25	4/4/0	0	1:50
11	25	4/0/0	0	1:50
12	25	4/4/1	25	1:50
13	25	4/4/0	0	<25
14	25	4/0/0	0	<25
15	22	5/0/0	0	<25
16	22	4/4/4	100	1:500
17	25	4/0/0	0	<25

* Number of grams of heart used for digestion and mouse bioassay.

† Number of mice inoculated/number of mice positive for *T. gondii*/number of mice that died from inoculation of heart tissue.

‡ Percentage of mice inoculated with that isolate that died.

§ Titers obtained in the MDAT for *T. gondii*.

|| Not applicable.

¶ No mice inoculated with this isolate died; however, mice had clinical signs of infection prior to necropsy.

Toxoplasma gondii was isolated from 8 (50%) of the 16 wild turkey hearts examined (Table 1). Six (75%) of the 8 isolates caused fatal infections in mice on primary isolation (Table 1). Prior to necropsy, mice in another group had clinical signs of weight loss and rough hair coats that were suggestive of *T. gondii* infection. Results of the modified direct agglutination tests demonstrated that 12 (71%) of the 17 samples had titers $\geq 1:50$ (Table 1). *Toxoplasma gondii* was isolated from the heart of 1 wild turkey that was negative at 1:25 in the MDAT (Table 1).

The prevalence of *T. gondii* infection in wild turkeys from Alabama in the present study is much higher than that reported by Burrige et al. (1979) for wild turkeys from Florida. Burrige et al. (1979) found no positive samples from the 20 wild turkeys they examined using an indirect hemagglutination test. However, negative serological reactions to *T. gondii* in these birds were probably a result of the serological test employed. Avian sera do not react as do mammalian sera in all serological tests (Frenkel, 1981), and tur-

keys experimentally infected with *T. gondii* do not develop titers or have low titers in the indirect hemagglutination test (Dubey et al., 1993). We used the MDAT to detect *T. gondii* antibodies in wild turkey sera because the MDAT has been validated in domestic turkeys (Dubey et al., 1993) and other wild birds (Kirkpatrick et al., 1990; Lindsay et al., 1991; Dubey et al., 1992).

In the present study, the serological prevalence of *T. gondii* was 71%, and the direct isolation prevalence was 50%. In one instance, *T. gondii* was isolated from the heart, but the serum/tissue fluid had a titer of <1:25 and was considered negative (Table 1). This may represent a recent infection in this bird, in which case it did not have sufficient time to mount a serological immune response. Alternatively, the bird may have had an antibody titer that was less than 1:25 and not detected. The lack of isolation of *T. gondii* from 4 wild turkeys that were MDAT-positive (Table 1) probably indicates that no tissue cysts were present in the heart but may have been present in other tissues. *Toxoplasma gondii* has been isolated from the brain, heart, breast muscles, and leg muscles of experimentally infected domestic turkeys (Drobeck et al., 1953; Dubey et al., 1993).

Ground-feeding birds, such as wild turkeys, probably acquire infection with *T. gondii* by ingesting sporulated oocysts from the soil. Feral cats (*Felis catus*) and bobcats (*Felis rufus*) are likely sources of oocysts of *T. gondii* for free-ranging wild turkeys.

The high prevalence of encysted *T. gondii* in wild turkeys indicates that these birds are potential sources of human infection. Freezing (-12.4°C for <1 min) and adequate cooking ($\geq 64^{\circ}\text{C}$ for 3.6 min) readily kill *T. gondii* tissue cysts in pork (Dubey et al., 1990; Kotula et al., 1991); therefore, common food preparation practices should kill most bradyzoites in tissue cysts in wild turkey tissue. Viscera from wild turkeys should not be fed to domestic cats. If turkeys are field-dressed, then the viscera should be buried or burned to prevent infection in wild felids or other animals.

We thank Natasha S. Rippey and Lisa C. Parsons for technical assistance. We thank Dr. J. P. Dubey, USDA, Zoonotic Diseases Laboratory, Beltsville, Maryland, for conducting the MDAT on turkey sera/tissue fluids. This study was supported in part by funds from the Department of Pathobiology, College of Veterinary Medicine, Auburn University.

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J. Helminthol. Soc. Wash.
61(1), 1994, pp. 117–121

Research Note

Seroprevalence of *Toxoplasma gondii* in Wild Mammals in Kansas

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ABSTRACT: Between 1989 and 1993, serum samples from 516 mammals in Kansas representing 17 species were examined for antibodies to *Toxoplasma gondii* using a modified direct-agglutination test. The overall prevalence was 84/516 (16%) mammals infected. When considering only animals where adequate sample sizes were available, the highest prevalences were found in raccoons (*Procyon lotor*), of which 14/20 (70%) were infected; white-tailed deer (*Odocoileus virginianus*), with 47/106 (44%) seropositive; and Virginia opossums (*Didelphis virginiana*), with a prevalence of 9/28 (32%). The seroprevalence in rodents and pronghorn antelopes was <8%.

KEY WORDS: *Toxoplasma gondii*, coccidia, survey, deer, raccoon, Kansas.

The intermediate host range of *Toxoplasma gondii* (Nicolle and Manceaux, 1908) Nicolle and Manceaux, 1909, is unusually wide for a coccidian, currently comprising >350 known species of vertebrates (Beyer and Poljansky, 1989). Felids, the definitive hosts, are thought to become infected principally by ingesting tissues of infected mammals and birds, and most cats are seropositive by weaning time (Dubey and Beatie, 1988).

Although numerous studies have examined the seroprevalence of antibodies to *T. gondii* in humans and wild and domestic animals, limited information exists concerning the seroprevalence of this coccidian in Kansas. Lindsay et al.

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